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Part I

PODIUM ABSTRACTS

1 3D Solitary Sequence Magnetic Resonance Imaging of the Dog Spine in Hansen Type I Intervertebral Disk Disease

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INTRODUCTION: Sampling perfection with application optimized contrasts using different flip angle evolutions (SPACE) is a T2-weighted three-dimensional MRI sequence. SPACE is potentially advantageous to standard MRI because it is a single acquisition faster, and technically easier technique with higher resolution and multi planar reconstruction capabilities. Hypothesis: SPACE is able to positively identify intervertebral disk disease with equal accuracy to T2-weighted spin echo imaging.

MATERIALS & METHODS: A double-blinded controlled study of dogs with an acute onset thoracolumbar myelopathy secondary to IVDD. All patients underwent magnetic resonance imaging of the T3–L3 spine using a 1.5T scanner. Images were obtained with the following sequences: standard T2–W sagittal and transverse, STIR dorsal, and SPACE. All dogs underwent hemilaminectomy based on the imaging findings. The standard T2-weighted and SPACE sequences were individually evaluated for site, side, and degree of, surgical recommendation and subjective confidence of imaging findings.

RESULTS: Twenty-three dogs met the inclusion criteria. There was statistically significant agreement for both site and side (p-value <0.001) between standard imaging and SPACE. There was complete agreement by all three researchers concerning site and side of disk herniation for 20/23 dogs, and partial agreement for the remaining 3. Compressive length ratio estimates by the two methods were similar, with more variability due to evaluators than differences between the two systems.

DISCUSSION/CONCLUSION: The MRI sequence SPACE was able to positively identify Hansen Type I Intervertebral disk disease in the canine thoracolumbar spine as confirmed via surgery with equal accuracy to standard T2-weighted imaging.

ACKNOWLEDGEMENT: No proprietary interest for this project.

2 Effects of Administration of Adipose-Derived Stromal Vascular Fraction and Platelet-Rich Plasma to Dogs with Osteoarthritis of the Hip Joints

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INTRODUCTION: The objective of this study was to evaluate effects of simultaneous intra-articular and IV injection of autologous adipose-derived stromal vascular fraction (SVF) and platelet-rich plasma (PRP) to dogs with osteoarthritis of the hip joints.

MATERIALS & METHODS: Twenty-two client-owned dogs were enrolled in the study. Dogs with osteoarthritis of the hip joints that caused signs of lameness or discomfort were characterized on the basis of results of orthopedic examination, goniometry, lameness score, the Canine Brief Pain Inventory (CBPI), a visual analogue scale, and results obtained by use of a pressure-

sensing walkway at week 0 (baseline). Dogs received a simultaneous intra-articular and IV injection of SVF and PRP or a placebo. Dogs were examined again 4, 8, 12, and 24 weeks after injection.

RESULTS: CBPI scores were significantly lower for the treatment group at week 24, compared with scores for the control group. Mean visual analogue scale score for the treatment group was significantly higher at week 0 than at weeks 4, 8, or 24. Dogs with baseline peak vertical force (PVF) in the lowest 25th percentile were compared, and the treatment group had a significantly higher PVF than did the control group. After the SVF-PRP injection, fewer dogs in the treated group than in the control group had lameness confirmed during examination.

DISCUSSION/CONCLUSION: For dogs with osteoarthritis of the hip joints treated with SVF and PRP, improvements in CBPI and PVF were evident at some time points, compared with results for the control group.

ACKNOWLEDGEMENT: MediVet Biologics donated equipment and materials to this study.

3 Factors Associated with Tibial Tuberosity Fracture after Tibial Plateau Leveling Osteotomy

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INTRODUCTION: The purpose of this study was to evaluate factors associated with tibial tuberosity fracture (TTF) following tibial plateau leveling osteotomy (TPLO) and to ascertain if a ratio of tuberosity measurements could be defined to provide a more broadly applicable recommendation for TPLO planning.

MATERIALS & METHODS: Radiographs of 1081 stifles having undergone TPLO were evaluated for cases of TTF. Seventeen variables were recorded. Two ratios were generated, the height of the tuberosity (TTH) over the width of the tibia at the most distal extent of the osteotomy (TW). The second ratio consisted of the width of the narrowest point of the tuberosity (TTW) over TW.

RESULTS: 10 (0.9%) of stifles experienced TTF. Tuberosities with the narrowest point below the level of insertion of the patellar ligament (PL) were 7.1 times more likely to fracture. Anti-rotational pins placed below the level of PL insertion were 10.2 times more likely to fracture. TTH/TW was not significant, but TTW/TW in the fracture group was significantly smaller (more narrow) than the control group.

DISCUSSION/CONCLUSION: Tuberosities narrowest distal to the PL insertion as well as distal insertion of anti-rotational pins may predispose to TTF. Preserving a tibial tuberosity which at the narrowest point is approximately 40% the width of the tibia at the base of the osteotomy site may help prevent TTF.

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4 Meniscal Kinematics through Automatic 3D Meniscus Model Extraction from MR Images: Validation and Application in Canine Stifles

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INTRODUCTION: Meniscal tears are known to be associated with structural progression of stifle osteoarthritis and commonly occur during twisting or flexing movements. The objectives of this study were to develop a meniscal study framework by investigating three-dimensional description of meniscal kinematics at four different positions, and developing an automatic image segmentation algorithm.



MATERIALS & METHODS: In this study, menisci from 16 cadaveric dog stifles were analyzed by 3T MR system with proton density sequence in the sagittal plane at four different weight-bearing knee positions. MRIs were manually segmented and kinematics were quantified using a new method. These manually segmented models were used to develop a statistical shape model, which was deformed to match the menisci in the MRI automatically. Our scheme was validated by comparison with manually segmented models in a leave-one-out paradigm.

RESULTS: Meniscal displacements were large, displacing 13.5mm and 13.7mm caudally on average for the lateral and medial menisci during flexion ($p=0.90$). The medial cranial horn and lateral caudal horns were the most mobile structures, showing average translations of 15.9mm ($p=0.371$) and 15.1mm ($p=0.019$), respectively. The automatically extracted meniscal models showed mean absolute surface distance errors of 0.43mm and 0.48mm, and average Dice overlap coefficients of 81.1% and 81.9% for medial and lateral meniscus, compared to manually segmented models.

DISCUSSION/CONCLUSION: A novel technique for automatic meniscal segmentation has now been validated, and preliminary studies of meniscal kinematics show mechanics of direct relevance to canine clinical care and the use of dog stifles as translational models for human medicine.

ACKNOWLEDGEMENT: Funded by UF opportunity fund.

5 In-Vivo Three-Dimensional Stifle Kinematics in Goats with Anterior Cruciate Ligament Deficiency

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INTRODUCTION: Goats are commonly used for translational ACL research. Reported deleterious effects of ACL transection in this species are inconsistent; thus the limitations of this model are poorly defined. Quantification of in vivo, 3D kinematics of ACL-deficient caprine stifles will improve insight regarding the value of caprine ACL research.

MATERIALS & METHODS: Fluoroscopic imaging of goat stifles ($n=6$) and force plating was performed before and at 2 weeks and 3 months after unilateral, arthroscopic ACL transection. Three-dimensional bone models were superimposed over fluoroscopic images. Relative alignment in 6 degrees of freedom was calculated and kinematics were compared as a percentage of gait cycle. Repeated measures ANOVA was used to compare mean kinematic parameters of the right stifle. $p<0.05$ was considered significant.

RESULTS: A significant reduction in peak vertical force occurred in the operated limb at 2 weeks post-ACL transection. Flexion/extension, axial rotation, and abduction/adduction were not significantly different from baseline. Anterior tibial translation was significantly increased during stance phase at 2 weeks and 3 months compared to baseline, with mean increases of 2.78 ± 0.78 mm and 4.39 ± 0.94 mm, respectively.

DISCUSSION/CONCLUSION: The magnitude of anterior tibial translation following ACL transection in this caprine model is similar to previous reports in humans. Lameness resolved by 3 months despite significant persistent kinematic abnormalities. The results of this study suggest that the caprine model is suitable for translational ACL research, as the kinematic consequences of ACL transection are representative of what has been observed with ACL deficiency in humans.

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6 Small Field of View 3T MRI to Determine Meniscal Volume in the Canine Stifle: An Ex-Vivo Study

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INTRODUCTION: MRI is a validated method to determine meniscal volume in human beings; however, the ability of MRI to accurately measure meniscal volume in dogs remains unknown. The objective of this study was to compare meniscal volume in normal canine menisci using small field of view 3T MRI and the Archimedes volume displacement method.

MATERIALS & METHODS: Seventeen canine cadaveric stifles were imaged with small field of view 3T MRI using proton density (PD) sequences. A single investigator performed meniscal segmentation for both medial and lateral menisci (34 total menisci) and determined meniscal volume using Mimics 16.0 (Materialise, Plymouth, MI). Menisci were dissected and volume was determined by the Archimedes method. Data were reported as mean \pm SD, 95% Confidence Intervals, and analyzed using Paired t-tests and Spearman correlation coefficients (significance $p<0.05$).

RESULTS: Meniscal volume for the 34 menisci was significantly greater using volume displacement (Mean: $784.0 \pm 237.0 \mu\text{l}^3$; 95%CI: $701.6-867.0 \mu\text{l}^3$) versus 3T MRI (Mean: $718.4 \pm 226.5 \mu\text{l}^3$; 95%CI: $639.4-797.5 \mu\text{l}^3$) ($p<0.0001$). Interestingly, there was a strong correlation between the two techniques ($R^2 = 0.9290$).

DISCUSSION/CONCLUSION: Although 3T MRI consistently underestimated meniscal volume, the strong correlation suggests a predictive relationship between the two techniques. Mathematical modelling is in progress to define this relationship in preparation for future meniscal tissue engineering studies.

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7 Quantitative MRI and Epic Micro CT for Assessing Cartilage Proteoglycan Content in the Canine Stifle

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INTRODUCTION: Delayed gadolinium-enhanced MRI of cartilage (dGEMRIC), T1rho MRI, and equilibrium partitioning of an ionic contrast (EPIC) via μCT have all been shown to correlate with cartilage proteoglycan concentrations in species other than the dog. The purpose of this study was to determine whether assessments of cartilage health using these methods would correlate with cartilage proteoglycan concentrations in canine stifles with induced osteochondral injury.

MATERIALS & METHODS: Twelve research beagles had 8 mm diameter osteochondral defects created in the medial femoral condyle of each stifle. Six months post injury 8 dogs had dGEMRIC and T1rho MRI imaging. All twelve dogs were humanely euthanized and proteoglycan concentration was assessed from cartilage biopsies taken from the medial and lateral femo-



ral condyles, tibial plateaus, the trochlea, and the patella. Following biopsy acquisition the stifles were assessed using EPIC-microCT. Correlations between the measured proteoglycan content and dGEMRIC values, T1 ρ values, and μ CT measures of cartilage thickness, volume, and attenuation were performed.

RESULTS: There were no statistically significant correlations between the proteoglycan composition and dGEMRIC values ($p>0.05$) or T1 ρ values ($p>0.05$). The proteoglycan concentration was correlated with μ CT determined assessments of cartilage thickness ($r=0.55$; $p<0.001$) and attenuation ($r = -0.51$, $p<0.0001$) with a trend toward correlation with μ CT assessment of cartilage volume ($p = 0.055$).

DISCUSSION/CONCLUSION: The quantitative MRI techniques used in this study (dGEMRIC and T1 ρ) need further optimization for canine application. EPIC microCT appears a valid method for assessing proteoglycan concentration post mortem.

ACKNOWLEDGEMENT: Lynn Reece, Lisa Reno, Ethan Karsted, and Kim Mason for technical assistance.

8 Comparison of Tibial Morphology in Normal and Cranial Cruciate Ligament Deficient Stifles of 18 Dogs

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INTRODUCTION: Our objective was to evaluate the morphological differences in the articular surface geometry of the tibia between the CCL insufficient and the contralateral normal stifle in dogs with unilateral CCL insufficiency using computed tomography (CT). We hypothesized that there would be greater asymmetry in condyle sphericity in the lateral vs. medial compartment in the CCL insufficient stifle.

MATERIALS & METHODS: Dogs with unilateral CCL insufficiency were recruited. CT of both hindlimbs was performed. CT Measurements were performed, including: medial and lateral radius of curvature of the tibial plateau subchondral surface (TPr), medial and lateral tibial plateau angle (TPA) and tibial plateau width (TPw). Parameters were compared between CCL insufficient and normal stifles using the two-sample paired t-test. Owners were contacted >1 year following CT scans, dogs with evidence of contralateral CCL disease were excluded.

RESULTS: Eighteen dogs met our inclusion criteria. Mean (\pm SD) age of the dogs was 83.6 (\pm 24.1 months) with a mean weight of 28.6 (\pm 7.0 kg). Mean follow-up time was 39.1 (\pm 24.2 months). Tibial variables associated with CCL insufficiency included a wider tibial plateau width ($p=0.0002$), larger medial TPr ($p=0.0016$); and larger medial TPr/lateral TPr ratio ($p=0.0002$). No other variables were found to be significantly different.

DISCUSSION/CONCLUSION: Our results demonstrate differences in proximal tibial morphology between CCL insufficient and normal stifles. While these changes may reflect underlying morphologic abnormalities leading to CCL insufficiency, it is possible that the differences observed were the result of osteoarthritic change.

ACKNOWLEDGEMENT: Financial support for this project was provided in part by the VOS Hohn-Johnson award.

9 Temporal and Regional Variation in Gene Expression, Histopathology and Biomechanical Properties in an Ovine Model of Flexor Tendinopathy

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INTRODUCTION: Flexor tendinopathy is a common condition with pathological changes distributed throughout the entire tendon. The aim of this study was to investigate the gene expression, histopathological and biomechanical changes that occur 8, 12 and 16 weeks after tendinopathy is induced.

MATERIALS & METHODS: Merino wethers (2 years old, $n=21$) underwent a partial transection of the hindlimb superficial digital flexor tendon (SDFT). Transected tendons and contralateral control tendons were harvested (7 sheep per time point) and regionally sampled for gene expression (real time PCR), histologic and biomechanical analyses at 8, 12 and 16 weeks postoperatively.

RESULTS: Surgical injury was associated with decreased gene expression for aggrecan, decorin, fibromodulin, tissue inhibitors of metalloproteinases (TIMPs 1, 2 and 3), collagen-I and collagen-II ($P < 0.05$). There were increases in expression for collagen-III, lumican and matrix metalloproteinase-13 ($P < 0.05$), which decreased over time but remained regionally high in comparison to controls ($P < 0.05$). TIMP3 increased over time ($P < 0.05$). Histologically, operated tendons had higher pathology scores than controls, especially adjacent to the injury ($P < 0.05$). Pathological changes in cellularity, vascularity, proteoglycan accumulation and collagen fiber alignment did not improve over time ($P < 0.05$).

DISCUSSION/CONCLUSION: The study improves understanding of the tendon environment post-injury, identifying a 'peak' of pathology that partially resolves over time. High localized collagen-III levels may reflect the production of tendon tissue with suboptimal biomechanical properties. Long-term studies evaluating tendon behavior post-injury are warranted to provide additional insights into tendon healing.

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10 MicroRNA-29a in Equine Tendinopathy

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INTRODUCTION: Tendinopathy in horses is characterized by over-expression of structurally inferior collagen 3 (col3) at the site of tendon injury. Recent work in humans has shown that this process is orchestrated through the injury-induced loss of microRNA-29a. The aim of this study was to assess the role of miR-29a in mediating col3 expression in an equine model of tendinopathy.

MATERIALS & METHODS: Thoroughbred horses ($n=8$), had collagenase-induced lesions in the superficial digital flexor tendon (SDFT). To establish tendon explant cultures, the SDFT was aseptically excised from the midmetacarpal region of both forelimbs of 3 horses. The impact of miR-29a was measured using quantitative-PCR, luciferase and Luminex cytokine multiplex assays.

RESULTS: Col3 transcript levels were elevated at all time points of the 16-week trial peaking at week 3, when col3 levels were 80-fold increased compared to uninjured controls. In contrast, miR-29a expression was significantly ($p<0.01$) down-regulated throughout all time points. Col3 luciferase assay confirmed that miR-29a directly targets the 3' untranslated regions of col3. Furthermore, introduction of synthetic miR-29a into cultured equine



tenocytes reduced col3 expression. Additionally miR-29a inhibition resulted in a significant increase in col3 expression indicating that miR-29a is not only actively regulating col3 but its loss is an important factor in the increase of col3 production observed in tendinopathy.

DISCUSSION/CONCLUSION: This work shows that injury-induced loss of miR-29a is responsible for the over-expression of col3 seen in tendinopathy.

ACKNOWLEDGEMENT: None.

11 In-Vivo Evaluation of Sedation on Acoustoelastography in Normal Equine Superficial Digital Flexor Tendons

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INTRODUCTION: Despite the high re-injury rate of superficial digital flexor tendon (SDFT) lesions in athletic horses, it remains difficult to determine the level of work that damaged tissue can sustain based on serial ultrasound examinations alone. Acoustoelastography (AEG) is a new ultrasound-based model that deduces stiffness gradient determining the pathology index (PI), which represents the rate of change in stiffness in relation to the change in strain. PI is obtained analyzing the changes in echo intensity observed in cine loops captured from gradually deforming tendon. Sedation is usually necessary for AEG examination and may affect PI. The purpose of this study was to assess the effect of sedation on AEG of normal equine SDFTs.

MATERIALS & METHODS: PI for the SDFT of 27 clinically normal horses were evaluated before and after sedation at levels 5, 10, 15 and 20cm DACB. Loading of the SDFT was achieved by lifting the contralateral forelimb during image acquisition. Repeated-measures ANOVA was used for statistical analysis.

RESULTS: PI after sedation was overall lower than before sedation. PI was significantly lower distally than proximally. Differences between individual horses accounted for the largest variance effect.

DISCUSSION/CONCLUSION: AEG is a helpful tool to evaluate tendon function; however this study shows that sedation may impact AEG. Clinically therefore AEG tendon evaluations should be consistent for each patient. This study had as limitation that only one sedation protocol was evaluated.

ACKNOWLEDGEMENT: R Vanderby Jr holds a patent on some aspects of the AEG technology. Funding sources – Merck-Merial Summers Scholars Program.

12 Muscle Injuries As the Primary Cause of Lameness in Fourteen Horses

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INTRODUCTION: The objective of this study was to describe clinical and ultrasonographic findings and outcome of horses having lameness primarily due to a muscle injury.

MATERIALS & METHODS: Medical records were reviewed for horses having a diagnosis of muscle injury following ultrasonographic evaluation. Cases were included only if horses had a lameness primarily attributable to muscle injury.

RESULTS: Fourteen horses were identified and all displayed lameness at presentation (grade 2/5–5/5 AAEP scale). Time between onset of lameness and presentation ranged from 6 hours to 6 weeks. Typical ultrasound findings included discontinuity of muscle fiber pattern, intermuscular fascial injury and diffuse/circumscribed hematomas. Eight cases involved a single muscle and 6 cases involved ≥ 2 muscles. Most horses survived to discharge (13/14), one was euthanized on presentation due to injury severity. One horse was euthanized 4 weeks after discharge due to development of contralateral limb laminitis. Of the remaining twelve cases, eleven were alive at long-term

follow up (8–90 months). One was euthanized for unrelated reasons 12 months after discharge. Recovery time ranged from 6–52 weeks (mean 19.8 weeks). Healing times were <16 weeks for single muscle injuries and 12–52 weeks for multiple muscle injuries. Three of the twelve long-term survivors had chronic lameness/stiffness preventing them from returning to their previous activity level. The remaining cases (9/14) returned to an equal or greater level of activity.

DISCUSSION/CONCLUSION: Based on this study, favorable long-term outcomes (9/14 = 64%) are expected in horses having muscle injuries as a primary cause of lameness.

ACKNOWLEDGEMENT: There was no proprietary interest or funding provided for this project.

13 Long-Term Outcome of Horses Treated with Lateral Digital Extensor Myotectomy for the Treatment of Conventional Stringhalt: 10 Horses (2002–2015)

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INTRODUCTION: The objective of this study is to report the outcome of horses having lateral digital extensor (LDE) myotectomy for the treatment of stringhalt in a current population of cases from the USA.

MATERIALS & METHODS: Medical records (2002–2015) from cases with a diagnosis of stringhalt that underwent LDE myotectomy were included. Horses were subdivided in 4 groups with follow up based on client conversations. Group 1 included horses with full improvement. Group 2 incorporated horses that had moderate improvement with mild residual hyperflexion. Group 3 had mild improvement with moderate residual hyperflexion. Group 4 incorporated horses that had no improvement. Follow-up included documenting complications from surgery, return to intended/athletic use and owner satisfaction.

RESULTS: Long-term follow up was obtained in 10/13 horses, ranging from 8 months to 10 years. Three of 10 (30%) horses had complete resolution (Group 1), 2/10 (20%) had moderate improvement (Group 2), 1/10 (10%) horse had full improvement on the left hind limb and mild improvement of the right hind (Group 3) and 4/10 (40%) horses had no improvement (Group 4). Six of 10 (60%) horses were performing athletically prior to developing stringhalt and 4/6 (66.7%) were able to return to their previous athletic function. No complications were reported and 7/10 (70%) of the owners were satisfied with surgery.

DISCUSSION/CONCLUSION: LDE myotectomy procedure remains the treatment of choice for conventional stringhalt. Goals for the surgery include improving the gait of the affected horse, improving athletic performance, and quality of life.

ACKNOWLEDGEMENT: There was no proprietary interest or funding provided for this project.

14 Regional Limb Perfusion of Gadolinium Contrast Medium in 26 Horses

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INTRODUCTION: The use of gadolinium contrast medium is common in small animals to better assess lesions identified with magnetic resonance imaging (MRI); however, its use is limited in equine patients due to the large volume required for systemic administration. The purpose of this study was to validate the feasibility of administering contrast medium in a lower dose via venous regional limb perfusion.

MATERIALS & METHODS: Twenty-six horses were prospectively enrolled and their distal limb was imaged with MRI before and after administration of



5mL of Magnevist (gadopentetate dimeglumine) diluted with 5mL of physiologic saline via a palmar digital vein with a tourniquet in place at the level of the mid aspect of the third metacarpal/metatarsal bone. Images were reviewed.

RESULTS: Twenty-five of 26 horses had adequate contrast enhancement of their distal limb. The lack of adequate contrast enhancement in one limb was likely associated with failure of the tourniquet. No adverse reactions were identified in any horse. Commonly affected structures of the equine distal limb were examined for normal and abnormal contrast enhancement.

DISCUSSION/CONCLUSION: Regional limb perfusion is a feasible method for administration of gadolinium contrast medium to the equine distal limb for MRI. The normal enhancement pattern of the equine distal limb is described. Contrast enhanced MRI of the distal equine limb helps to further characterize lesions identified with pre-contrast images, including adhesions and deep digital flexor tendinopathy. Contrast enhanced MRI also helps identify additional lesions, such as neovascularization.

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15 Modeling Exercise and Humeral Fracture

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INTRODUCTION: Periods of reduced exercise preceding a return to high-speed exercise are associated with greater risk for humeral fracture in Thoroughbred racehorses. We hypothesized that length of layup and distribution of high speed activities affect humeral damage and thus fracture risk. Computational models examined the humeral response to various training programs at the site of stress fracture.

MATERIALS & METHODS: A finite element model of an equine humerus was used to simulate joint contact and muscle forces for three phases of stance (fore, mid, and aft) for walk, slow gallop (10 m/s), and fast gallop (17 m/s). Bone remodeling (BMUs) was governed by a mechanical stimulus that was dependent on strain and number of loading cycles for n gait loading conditions. BMUs removed bone in response to damage and to mechanical disuse. Bone damage was compared for different lengths of layup, and between one high-speed gallop or two split gallops a week.

RESULTS: Model predicted humeral damage was lower after return to training after longer layup and with high-speed exercise split over two days a week.

DISCUSSION/CONCLUSION: Model damage levels are consistent with epidemiologic data. Modified training may reduce humeral stress fractures.

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16 Measurement of Shoulder Abduction Angles in Dogs. An Ex-Vivo Study of Accuracy and Precision

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INTRODUCTION: Insufficiency of the medial glenohumeral ligaments results in increased shoulder abduction angles. To date, accurate and repeatable modalities for diagnosing medial shoulder instability (MSI) remain elusive. Although a non-invasive method for evaluating the integrity of medial shoulder support structures, the accuracy and precision of the shoulder abduction study test remains unknown.

MATERIALS & METHODS: Shoulder abduction angles were measured by three observers in 10, medium-to-large sized dog cadavers, before and after shoulder arthroscopy. One shoulder in each cadaver was randomly assigned to have the medial glenohumeral ligaments and the subscapularis tendon transected. Shoulder abduction angles were also measured using fluoroscopic images acquired in the cranio-caudal plane, as the shoulder was abducted. Both intra-observer (Interclass Correlation Coefficient) and inter-observer repeatability (Paired t-test) were assessed.

RESULTS: The interclass correlation coefficient (95% CI) was 0.244 (-0.022, 0.546) and 0.523 (0.264–0.755) for the pre and post-treatment categories, respectively. The mean difference (95% CI, p-value) between the first and second examinations was 4.933° (2.35–7.52, 0.002) for Observer 1; 9.93° (4.01–15.87, 0.004) for Observer 2 and 5.90° (2.12–9.59, 0.006) for Observer 3. There was also a significant difference in measurements when comparing fluoroscopic angles to those measured by the three observers.

DISCUSSION/CONCLUSION: Our results suggest that the shoulder abduction test has poor inter- and intra-observer repeatability. Significant differences were also found when abduction results were compared with the measurements obtained via fluoroscopy, for all three observers. These findings suggest that, in isolation, the shoulder abduction test should not be used as a definitive test for diagnosing MSI.

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17 The Association of Post-Operative Rehabilitation with Outcome in Dogs with Intervertebral Disk Herniation

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INTRODUCTION: There is limited data on the effect of post-operative interventions on outcome in dogs undergoing surgery for IVDH. The purpose of this study was to evaluate the effects of post-operative in-house rehabilitation in dogs undergoing surgery for IVDH. Our hypothesis was that post-operative rehabilitation would shorten recovery time, improve neurologic recovery, and limit complications.

MATERIALS & METHODS: Retrospective cohort study. Medical records of dogs (n=248) that underwent a single site, thoracolumbar, hemilaminectomy for IVDH were reviewed and characterized into two groups based on whether or not an in-house rehabilitation program was performed post-operatively. All dogs were non-ambulatory prior to surgery, and pre-operative and sequential post-operative Modified Frankel Scores were assigned. Time to ambulation, time to normal conscious proprioception, final Modified Frankel Score, and complications were compared between the groups.

RESULTS: Significantly more dogs returned to full neurologic function (final MFS of Grade 5) in the rehab group (33%) compared to the control group (9%). The estimated median time to return of normal conscious proprioception and median time to return of ambulation were significantly shorter in the control group (42 days and 14 days, respectively) compared to the rehab group (49 days and 28 days, respectively). The complication rate was significantly higher in the control group (29%) compared with the rehab group (16%).

DISCUSSION/CONCLUSION: In-house rehabilitation should be considered a part of the post-operative management in dogs recovering from surgical treatment of IVDH to improve neurologic function and reduce post-operative complications.

ACKNOWLEDGEMENT: No proprietary interest or funding was provided.



18 A Comparison of Sham Versus Actual Low Level Light Therapy (LLLT) in Treatment of Canine Elbow Osteoarthritis Pain and Lameness

Andrea Looney¹; Janice Huntingford²; Lauren Blaese³; Sabine Mann⁴; Joseph J. Wakshlag⁵

¹Anesthesia and Pain Management Services, Ethos Veterinary Health/Massachusetts Veterinary Referral Hospital, Woburn, MA, ²Essex Animal Hospital, Essex Ontario, ON, Canada, ³Ethos Veterinary Health/Massachusetts Veterinary Referral Hospital, Woburn, MA, ⁴Cornell University College of Veterinary Medicine, Population Medicine and Diagnostic Sciences, Ithaca, NY, ⁵College of Veterinary Medicine, Cornell University, Ithaca, NY

INTRODUCTION: The effect of photobiomodulation (LLLT) or sham light therapy on degenerative joint pain was studied in 20 dogs with natural occurring elbow osteoarthritis.

MATERIALS & METHODS: Dogs (n=20) were randomly assigned to receive either LLLT (group LLLT, n=11) or sham (group S, n=9) treatment, to elbows for 6 weeks. Clinician Lameness score, Helsinki Chronic Pain Index by blinded owner, and NSAID (nonsteroidal anti-inflammatory) dose were recorded pre and post treatment

RESULTS: A NSAID dose reduction was possible in 9/11 dogs in the LLLT group, and in 0/9 of group S dogs (P = 0.0003). There was greater improvement in lameness score post course of light therapy in the LLLT vs S group (p=0.001). A greater reduction in score was detected in 9/11 of the Helsinki Pain Score parameters in group LLLT (P<0.05).

DISCUSSION/CONCLUSION: Regular LLLT at 10–20J/cm²/joint for 6 weeks was successful in improving lameness score and owner perceived pain scores, and in allowing a reduction in NSAID dose in canine patients with elbow osteoarthritis

ACKNOWLEDGEMENT: This study was funded by a grant from the Waltham Foundation. Equipment was provided by Companion Therapy by Lite Cure. Neither supporter played a role in study design, data collection or analysis, and abstract submission.

19 Preclinical Evaluation of ^{117m}Sn Colloid as a Radiosynoviorthesis Agent for Treatment of Canine Elbow Joint Osteoarthrosis

J M Donecker¹; J C Lattimer²; K A Selting³; J M Lunceford²; J R Holland²; J Simon³

¹Convetra Inc., The Woodlands, TX, ²University of Missouri Veterinary Medical Teaching Hospital, Columbia, MO, ³IsoTherapeutics Group, LLC, Angleton, TX

INTRODUCTION: Osteoarthrosis, the most common disorder of the canine elbow joint, can be difficult to treat with conventional therapies. Radiosynoviorthesis (RSO) has been used successfully to treat various types of arthritis in humans. To determine the safety of ^{117m}Sn for RSO in dogs, we evaluated its effects after intra-articular injection in normal canine elbow joints.

MATERIALS & METHODS: Five young adult dogs were used. With the dog under general anesthesia, synovial fluid for cytology was aseptically collected from one elbow joint, followed by intra-articular injection of 2.5 mCi of ^{117m}Sn colloid. Data obtained pre-injection and 42 days post-injection included CBC, serum chemistries, urinalyses, radiographs and MRI/PET (¹⁸FDG) scans of both elbows. Urine, feces and blood samples were evaluated for radioactivity for 5 days post-injection along with radiation dosimetry. Each dog was euthanized 43 days post-injection and a complete necropsy performed.

RESULTS: Greater than 99.8% of ^{117m}Sn was retained in the injected joint at 42 days post-injection. Radiation output levels were below patient release levels on day 1 post-injection. The only change from baseline studies was a slight decrease in joint fluid volume in the injected joint on the MRI images.

DISCUSSION/CONCLUSION: Injection of ^{117m}Sn colloid into normal canine elbow joints produced no adverse effects and was well-contained within

the joint. The radiation dose to the whole body and major organs was minimal. These findings indicate that ^{117m}Sn colloid has potential as a safe agent for RSO in dogs and other domestic animals.

ACKNOWLEDGEMENT: This study was sponsored by Convetra Inc. (J. Donecker, Chief Veterinary Officer).

20 Minimally Invasive Sacroiliac Lag Screw Fixation Using a Dedicated Novel Instrument System – Apparatus and Technique Description

Loïc M. Déjardin¹; Albane H Fauron¹; Laurent P. Guiot²; Reunan P. Guillou²

¹Department of Small Animal Clinical Sciences, College of Veterinary Medicine, Michigan State University, East Lansing, MI, ²Department of Veterinary Clinical Sciences, College of Veterinary Medicine, The Ohio State University, Dublin, OH

A novel instrumentation and surgical technique were devised to optimize screw placement while reducing radiation exposure during minimally invasive osteosynthesis (MIO) via lag screw fixation of sacroiliac luxation/fractures (SIL/F)

The SIL instrument system (SILIS) consists of 1) *two variable friction 6-axes arms* dedicated to either reduction or fixation, 2) a *reduction handle*, or “joystick”, and 3) a *modified drill guide*.

A joystick inserted in the ischial tuberosity is used as a reduction aid. Once adequate reduction has been fluoroscopically confirmed, the joystick is secured to its assigned arm. Fluoroscopic sacral body location is facilitated by moving a washer above the skin. A modified drill guide secured to the fixation arm is inserted through a stab incision and manipulated until it appears as a circle over the sacral body. Sacral pilot and iliac glide holes are drilled prior to lag fixation and skin closure.

Subjective evaluation of screw placement and SIL/F reduction is performed on orthogonal pelvic radiographs. Objective measurements of sacral screw orientation and sacral purchase are obtained on CT MPR images.

As 6-axis arms provide stable SIL/F reduction and drill guide alignment throughout surgery, surgical personnel may step away from the patient during fluoroscopic imaging which reduces radiation exposure. Additionally, rigidly securing the drill guide to a dedicated arm virtually eliminates aiming bias during drilling.

Early SILIS technique evaluation showed accurate sacral screw placement and purchase in all cases, supporting the findings of a recent cadaveric study that demonstrated the superiority of MIO treatment of SIL/F compared to ORIF.

ACKNOWLEDGEMENT: No proprietary interest or funding was provided for this project.

21 Creating a Research Animal Fracture Models Database

Carl A. Kirker-Head¹; Kevin M Connolly¹; Vanessa Rubin¹; Abby Brisbois¹; Therese McNamee¹; Brian Ingle²

¹Clinical Sciences, Cummings School of Veterinary Medicine at Tufts University, North Grafton, MA, ²AO Foundation, 7270 Davos, Switzerland

INTRODUCTION: Research animal fracture models help us understand the biological process and develop new and improved interventions. Many, however, fail to meet critical scientific review. Also, no current search engines focus on fractures and relevant publications may be overlooked or reported out of context. Here we describe the development of a new fracture model database that will better inform investigators exploring this focus.

MATERIALS & METHODS: In vivo research animal fracture models were identified using existing databases and textbooks. Citations were uploaded into the new database created on a SharePoint 2010 platform. Publications were additionally classified by fracture model, species, breed, sex, age, weight, study focus, evaluation methods, dataset size, anesthesia/analgesia and euthanasia techniques, study duration and complications. Publications were also scored for clinical translation, animal welfare, and repeatability.



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25 Mechanical Comparison of a Type II External Skeletal Fixator and Locking Compression Plate in a Fracture Gap Model

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INTRODUCTION: The purpose of this study was to compare the stiffness of a Type II external skeletal fixator (ESF) to a 3.5 mm locking compression plate (LCP) in mediolateral, and craniocaudal bending in comminuted fracture model. Our hypothesis was that the Type II ESF would demonstrate comparable stiffness to the LCP.

MATERIALS & METHODS: A 40 mm fracture gap was used to mimic a comminuted fracture. The locking compression plate (LCP) construct consisted of a 12 hole 3.5 mm LCP with three 3.5 mm bicortical locking screws placed per fragment. The Type II ESF construct consisted of 3 proximal full fixation pins (Centerface®) and 3 distal full fixation pins in the mediolateral plane, and 2 carbon fiber connecting rods. Five constructs of each were tested in non-destructive mediolateral and craniocaudal bending and axial compression. Stiffness was determined from the slope of the linear elastic portion of the load-displacement curves.

RESULTS: There was a significant difference between the stiffnesses of the Type II ESF and the LCP in all modes of loading except craniocaudal bending. The Type II ESF was significantly more stiff in mediolateral bending versus the LCP.

DISCUSSION/CONCLUSION: A Type II ESF withstands higher mediolateral bending forces compared to an LCP in a comminuted fracture model. This knowledge can augment a clinician's ability to choose an appropriate fixation method for a comminuted fracture.

ACKNOWLEDGEMENT: LCPs, screws and associated instrumentation were donated by Synthes Vet. This work was supported through funding by the VA-MD College of Veterinary Medicine.

26 Neurectomy of the Deep Branch of the Lateral Palmar Nerve As a Treatment for Chronic Equine Forelimb Proximal Suspensory Desmitis: Long Term Follow up on 20 Cases

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INTRODUCTION: A small proportion of forelimb proximal suspensory desmitis (PSD) cases have persistent lameness despite appropriate management. The hypothesis behind this treatment was that there may be persistent pain and lameness despite adequate healing of the ligament, in a similar fashion to hindlimb PSD, and that focal neurectomy would be effective in returning these cases to soundness without recurrent injury of the ligament.

MATERIALS & METHODS: Inclusion criteria for clinical cases to undergo surgery were a positive response to blocking the deep branch of the lateral palmar nerve (DBLPN), and an ultrasonographic appearance consistent with chronic desmitis without loss of structural integrity. Under general anesthesia, a 3–4cm skin and fascial incision was made distal to the accessory-metacarpal ligament. Three cm of the DBLPN was removed using a guillotine technique. The fascia and skin were closed routinely and the horses rested for a minimum of 4 weeks before return to exercise

RESULTS: The median duration of lameness prior to surgery was 10 months. Eighteen cases have long-term follow-up. Fifteen/18 (83%) were in full work at 12 months post-surgery; and 10/12 (83%) still in full work at two

years. None suffered severe suspensory breakdown post-operatively, or evidence of neuroma formation.

DISCUSSION/CONCLUSION: This appears to be an effective treatment for chronic forelimb proximal suspensory desmitis, in cases deemed to have minimal loss of structural integrity.

ACKNOWLEDGEMENT: Alicia Fuller and Carly Briggs for data collection. There was no proprietary interest or funding provided for this project

27 Outcome of Horses Undergoing Navicular Bursotomy for the Treatment of Contaminated or Septic Navicular Bursitis: 19 Cases (2002–2016)

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INTRODUCTION: Contaminated or septic navicular bursitis is reported to have guarded prognosis after the navicular bursotomy procedure. In our experience, the navicular bursotomy procedure, combined with local antimicrobial perfusion techniques, can provide a better prognosis for survival than previously reported. The objective of this retrospective study is to report the outcome of horses that underwent navicular bursotomy for the treatment of contaminated or septic navicular bursitis.

MATERIALS & METHODS: Medical records (2002–2016) were reviewed for horses that underwent navicular bursotomy. Follow-up information was obtained by contact with owners. Horse outcome was subdivided into 2 groups. A successful outcome (Group 1) was assigned to horses that were able to return to the same level of use and performance as before surgery. A satisfactory outcome (Group 2) was assigned to horses that survived but did not return to their previous function or level of performance.

RESULTS: All 19 cases (100%) survived to discharge. Follow up was obtained 4 months to 12.75 years after surgery. Sixteen (16) cases were able to return to their previous level of use (84.2%) and three (3) cases were able to return to a lower level of performance or were pasture sound (15.8%). All nineteen (19) owners were satisfied with the outcome at follow up.

DISCUSSION/CONCLUSION: A navicular bursotomy procedure is an effective treatment for contaminated or septic navicular bursitis. This technique, combined with local antimicrobial perfusion techniques, results in an excellent prognosis for survival and good prognosis for return to intended use, including athleticism.

ACKNOWLEDGEMENT: There was no proprietary interest or funding provided for this project.

28 Cell-Free DNA Concentrations in Equine Synovial Fluid of Healthy Young Horses Following Induction of Osteoarthritis of the Carpus and Treadmill Exercise

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¹Massey University, Palmerston North, New Zealand, ²University of Waterloo, Waterloo, ON, Canada, ³Clinical Sciences, Colorado State University, Fort Collins, CO

INTRODUCTION: Only limited information is available on equine plasma cell-free DNA (cfDNA) while there is no published information on equine synovial fluid (SF) cfDNA. Our objectives were to report baseline concentrations of SF cfDNA in untrained young healthy Thoroughbreds, and to determine the effects of induced osteoarthritis (OA) and exercise on SF cfDNA levels over time.

MATERIALS & METHODS: On Day 0 SF was collected from both middle carpal joints of 17 Thoroughbred fillies. Subsequently OA was surgically induced in one middle carpal joint in 9 fillies, while 8 fillies underwent a Sham operation. All horses commenced a 7-week treadmill exercise program on



Day 14. SF was collected weekly until Day 63, and cfDNA concentrations measured using a fluorimetric technique.

RESULTS: The median SF cfDNA on Day 0 was 504 µg/L (IQR= 236). SF cfDNA levels in OA joints were significantly higher than at Day 0 and were also significantly higher than Sham joints on days 28 and 63. SF cfDNA concentrations were significantly higher in Sham joints on days 7, 14, 21, 35, 42 and 63 compared to Day 0 values.

DISCUSSION/CONCLUSION: The effects of induced OA on SF cfDNA were sustained when compared with controls and exercise alone was not responsible for the changes observed. Validation of SF cfDNA use with naturally occurring disease cases is necessary before any clinical recommendations can be made. This study is the first to report equine baseline SF cfDNA concentrations, but its significance with respect to focal orthopedic trauma is unknown.

ACKNOWLEDGEMENT: Equine Trust and Lewis Fitch Veterinary Research Fund.

29 Use of a Bipolar Radiofrequency Device to Remove Proximal Sagittal Ridge Fragments of the Third Metacarpus/Metatarsus in Thoroughbred Yearlings

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INTRODUCTION: Elective arthroscopy is often the treatment of choice to remove osteochondral fragments diagnosed on pre-sale radiographs of Thoroughbred yearlings to reduce the incidence of secondary osteoarthritis leading to increased sale value. Osteochondritis dissecans (OCD) lesions from the proximal sagittal ridge (PSR) of the third metacarpus/metatarsus may often be embedded in the synovial plica making complete removal difficult. The purpose of this study is to describe an arthroscopic technique that uses a bipolar radiofrequency device to precisely dissect PSR fragments from those soft tissue attachments to facilitate complete removal, minimize inadvertent tissue trauma, and decrease radiation exposure.

MATERIALS & METHODS: Ten Thoroughbred yearlings were admitted for removal of PSR fragments following pre-sale examination including digital radiography as well as conformation and lameness evaluation. Once visualized arthroscopically, a bipolar radiofrequency device was used to dissect the soft tissue attachments prior to fragment removal that was confirmed via intra-operative radiographs.

RESULTS: The use of a radiofrequency probe facilitated accurate dissection and reduced the number of intra-operative radiographs to confirm fragment removal. Post-operative evaluations of the affected joints had no significant radiographic abnormalities and were grossly normal with no increased synovial effusion or incisional complications. All horses were subsequently sold successfully at public auction.

DISCUSSION/CONCLUSION: Arthroscopic removal of PSR fragments using a bipolar radiofrequency device proved to be an effective technique by reducing tissue trauma, radiation exposure and perceived anesthetic time with no adverse clinical or radiographic findings of the joint.

ACKNOWLEDGEMENT: There was no proprietary interest or funding provided for this project.

30 Biomechanical Comparison of LCP Fixation to a Novel Pedicle Screw External Fixation for Mandibular Fracture Repair

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INTRODUCTION: The purpose of this study was to determine the biomechanical properties of a novel pedicle screw external fixator system for

mandibular fracture repair and compare it to Locking Compression Plates (LCP). It was hypothesized that the pedicle screw system would be a simple and intuitive fracture repair technique requiring less soft tissue trauma, limited risk to tooth roots, and providing adequate stability compared to the LCP configuration and other clinically relevant repair techniques.

MATERIALS & METHODS: Twelve mandibles were chosen at random and stabilized with either the locking compression plate (LCP) or the pedicle screws, rod, and interdental wires (6 mandibles/treatment group). They were then tested in a single cycle to failure test and the failure point and stiffness were calculated.

RESULTS: Biomechanically, the pedicle screw construct was weaker and not as stiff as the LCP plate. Compared to a previous study, 1) the LCP construct had a higher failure point than the DCP construct; 2) the failure point of the pedicle screw construct was slightly lower than the DCP construct but still higher than a type II external fixator (EF), type II external fixator with interdental wires (EFW), and intraoral acrylic splint with interdental wires (ISW); and 3) stiffness was much greater for both the LCP and pedicle screw construct compared DCP, EF, EFW and ISW.

DISCUSSION/CONCLUSION: From the biomechanical data and clinical considerations, a polyaxial pedicle screw external fixator system offers a biomechanically comparable option that is simple, minimally invasive, and clinically applicable for mandibular fixation.

ACKNOWLEDGEMENT: There was no proprietary interest for this project.

31 Effects of Loading and Lag Screw Reduction on Type III Distal Phalangeal Fracture Stability

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INTRODUCTION: Type III distal phalangeal fractures in horses can be treated conservatively or surgically. Intuitively, fracture reduction should improve repair and mitigate secondary arthritis; however, retrospective outcomes do not support this. This study was carried out to determine the effects of lag screw stabilization and loading on these fractures.

MATERIALS & METHODS: Type III fractures were created in 12 paired digits. Fractures were reduced with 4.5 or 5.5mm cortical screws in lag fashion, or left as non-reduced controls. Fracture reduction was assessed by comparing pre- and post-reduction fracture gap measurements. Effects of incremental compressive loading and terminal unloading were measured from serial fluoroscopic images.

RESULTS: 5.5mm screws significantly reduced fracture gaps. 4.5mm screws did not. Under load, articular fracture gaps in all groups were compressed, and were closed at 540 Kg loads. Distally, loading increased fracture gaps in 'Control' and '4.5mm' groups. Reduction with 5.5mm screws significantly constrained distal expansion. After loading was removed, articular fracture gaps in Controls were wider than the residual gaps in reduced phalanges, although not significantly.

DISCUSSION/CONCLUSION: Only 5.5mm screws reliably reduced the fracture gaps, and were also more effective than 4.5mm screws in stabilizing distal fracture gaps under load. Fluoroscopic images clearly showed that loading the articular surface compressed the fracture gap, with complementary expansion of the fracture gap distally. These findings explain the success of conservative management of these fractures, although lag screw fixation does prevent cyclical fracture compression-expansion.

ACKNOWLEDGEMENT: There are no funding sources, proprietary interests or conflicts to report.



32 Effect of Carprofen Usage on Platelet Activation in Canine Platelet-Rich Plasma

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INTRODUCTION: It is unclear if use of non-steroidal anti-inflammatory drugs (NSAIDs) leading up to the time of platelet-rich plasma (PRP) preparation mitigates platelet activation and the potential efficacy of PRP. The purpose of this study was to evaluate the effect of carprofen on platelet activation in canine PRP.

MATERIALS & METHODS: Nine healthy dogs underwent PRP preparation. They were then administered carprofen (4.4mg/kg SID) for 7 days and PRP preparation repeated. Carprofen was continued for 4 more days, discontinued for 3 days, and PRP preparation performed a third time. All PRP samples were divided into two aliquots immediately following preparation and one aliquot was not activated and the other was activated with human gamma thrombin. All samples were then assessed with flow cytometry for expression of CD62P and platelet bound fibrinogen. The percentage of platelets positive for CD62P or CAP1 was evaluated using a one-way repeated measures analysis.

RESULTS: Fewer than 5% of platelets were positive for CD62P expression or platelet bound fibrinogen in all aliquots of PRP that had not been activated. Up to 100% of platelets in aliquots that were activated with human gamma thrombin were positive for CD62P or platelet bound fibrinogen. Carprofen administration had no effect ($p>0.05$) on percentage of platelets positive for CD62P or CAP1 in activated or unactivated PRP samples.

DISCUSSION/CONCLUSION: These results that indicate that use of carprofen does not preclude platelets in this canine PRP from being activated.

ACKNOWLEDGEMENT: Material support was provided by Arthrex Vet Systems.

33 Porous Poly(ethylene glycol) Diacrylate (PEG-DA) Hydrogels As Scaffolds for Articular Cartilage Tissue Engineering

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INTRODUCTION: A solvent-induced phase separation/solvent-casted particulate-leaching system (SIPS/SCPL) method was developed to generate (polyethylene glycol) (PEG) hydrogels containing a series of interconnecting pores to improve cell migration, proliferation, and tissue ingrowth.

MATERIALS & METHODS: Comparing conventional and SIPS/SCPL hydrogels, canine multipotent stromal cells (MSCs) were evaluated over 21 days for attachment using electron microscopy, cytotoxicity with a lactate dehydrogenase (LDH) assay and live/dead staining, proliferation using DNA quantification, and mechanical properties via dynamic mechanical analysis. Additionally, hydrogels were implanted subcutaneously or within femoral trochlea of rats and evaluated for biocompatibility at 21 days using histology. Data were reported as mean \pm SD and compared using ANOVA or Fisher's exact test. Significance was established at $p<0.05$.

RESULTS: MSCs cultured on SIPS/SCPL hydrogels exhibited attachment, spreading, and proliferation, whereas conventional hydrogels induced marked toxicity and lack of proliferation. SIPS/SCPL cultured MSCs expressed significantly lower LDH levels ($p<0.0001$) while cell number significantly

increased over time ($p<0.0001$). Storage modulus of SIPS/SCPL hydrogels increased significantly at 21 days for medium ($p<0.05$) and large ($p<0.001$) pore sizes. SIPS/SCPL hydrogels exhibited biocompatibility in vivo, as demonstrated by an acceptable inflammatory response, appropriate connective tissue ingrowth, neovascularization, and formation of woven bone adjacent to and within scaffolds ($p<0.0001$).

DISCUSSION/CONCLUSION: SIPS/SCPL hydrogels are promising scaffolds for articular cartilage tissue engineering strategies.

ACKNOWLEDGEMENT: This work was funded by the AKC-CHF.

34 In Vivo Re-Animation of Decellularized Skeletal Muscle Scaffold Following Volumetric Muscle Loss

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INTRODUCTION: Tumor excision and trauma often result in large muscular defects that cannot be completely repaired. We hypothesized that decellularized skeletal muscle scaffolds (DMS) seeded with skeletal muscle derived mesenchymal stromal cells (SMdMSCs) will integrate with minimal inflammation and have improved function when compared with muscles with no implant or those reconstructed with DMS alone.

MATERIALS & METHODS: 27 rats had a volumetric muscle defect created in the left anterior tibialis muscle (AT) and were randomly assigned to 3 groups: 1) empty defect, 2) DMS, and 3) DMS seeded with 1×10^6 Luc+SMdMSCs. At 16 weeks, peak isometric tetanic force (P_0) of the operated and non-operated AT was measured. The mean percent P_0 difference between operated and control AT and the mean histologic score of inflammation and scaffold integration were calculated and compared between groups ($p<0.05$).

RESULTS: Empty defect rats recovered 66% of normal P_0 at 4 months, DMS rats recovered 73% of normal P_0 and DMS/Luc+SMdMSCs treated rats recovered 97% of normal P_0 . A statistical difference between groups was not achieved ($p>0.05$). The DMS/Luc+SMdMSC group had a significantly greater mean histologic score of inflammation and integration as compared to the empty defect group ($p<0.05$).

DISCUSSION/CONCLUSION: The improved functional outcome for the DMS/Luc+SMdMSC group and the significantly greater histologic score of scaffold integration indicates that reanimation of decellularized muscle scaffolds has therapeutic promise for functional reconstruction of volumetric muscle loss.

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35 Exposure to Donor Bone Protein Induces Tolerance in Massive Cortical Allograft Recipients

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INTRODUCTION: Fresh-frozen cadaveric allografts are utilized to reconstruct large bone defects. Low-grade graft vs. host responses have been well-documented in cortical allograft recipients and immunosuppression results in superior healing. We previously showed that exposure to a vaccine comprised of donor bone proteins resulted in increased new bone formation. The objective of this study was to understand the immunomodulatory mechanism responsible for the improved healing.

MATERIALS & METHODS: A 5mm femoral defect was created in C57BL/6 mice ($n=40$) and reconstructed using a cortical allograft from a Balb/C donor ($n=30$) or a cortical isograft ($n=10$). Each group was divided as follows: allograft+ saline x4 ($n=10$), autograft+ saline x4 ($n=10$), allograft+ donor bone vaccine x4 ($n=10$) or allograft+ isograft bone vaccine x4 ($n=10$). Vaccines



were given 1 week prior to limb reconstruction, and on post-operative days 7, 21, and 35. Mice were euthanized on day 42. Recipient spleens were collected and exposed to allogenic bone protein or isogenic bone protein. T-cell response was assessed via flow cytometry. Graft integration was assessed via micro-CT.

RESULTS: T-cell response to donor bone was suppressed following bone protein vaccination. Micro-CT showed improved bone healing in allograft+donor bone vaccine mice as compared with recipients of allograft alone.

DISCUSSION/CONCLUSION: Poor allograft incorporation can be partially attributed to T-cell responses against graft antigens. T-cell response is modulated following exposure to a vaccine comprised of donor bone protein resulting in better overall bone healing. Vaccination with donor bone antigens may yield a promising immunotherapeutic to improve allograft integration.

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36 Comparison of Anabolic Growth Factor Elution from Thrombin/Calcium Chloride Activated Platelet-Rich Fibrin Gels and Platelet-Rich Chondroitin Sulfate Glycosaminoglycan Gels

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INTRODUCTION: Platelet-rich plasma (PRP) is an autologous blood product that is a common therapeutic in the treatment of musculoskeletal injuries, but efficacy remains unclear and clinical results vary. PRP's regenerative potential is thought to lie in its release of anabolic growth factors. Therefore, a sustained release administration method may improve efficacy in vivo.

MATERIALS & METHODS: PRP was made from eight healthy dogs using a commercially available device. Each PRP preparation was split into 2 aliquots: one activated with bovine thrombin/CaCl₂ to form a platelet-rich fibrin (PRF) gel, the other rehydrated a lyophilized chondroitin sulfate-glycosaminoglycan (CS-GAG) gel. Both gels were incubated in media for 7 days. Media was aspirated, saved, and replaced at 1, 3, 5, and 7 days. Media samples were frozen until assayed for TGF- β 1 concentrations by ELISA.

RESULTS: There was no significant difference in TGF- β 1 elution from the two gels at day 1. However, there was up to a 256% greater concentration of TGF- β 1 eluted from the CS-GAG gels compared to the PRF gels on days 3, 5, and 7 ($p < 0.05$). Significantly more TGF- β 1 was released from the CS-GAGs on days 5 and 7 compared to day 3.

DISCUSSION/CONCLUSION: One possible explanation for lack of PRP efficacy may be that the delivery of anabolic growth factors is transient. Use of negatively charged CS-GAG hydrogels may improve PRP efficacy by increasing the duration of anabolic growth factor delivery.

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37 Comparison of Various Freezing Medium Conditions on the Phenotype of Canine-Adipose Derived Stromal Cells

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INTRODUCTION: Mesenchymal stem cells (MSC) are cryopreserved for future treatments. Cryopreservation induces cell injury, altering viability and phenotype, requiring cryoprotective agents. Comparison of cryoprotectives are not reported in canine MSCs. Our objective was to compare cryo-

protective effects on viability, differentiation, and cytokines expression profiles of canine MSCs. Our hypothesis was that no significant difference exists.

MATERIALS & METHODS: Canine adipose-derived MSCs from four donors were placed in six cryoprotectant medium conditions: fetal bovine serum (FBS), canine serum (CS), trehalose, dimethyl sulfoxide (DMSO)+FBS, DMSO+CS, or DMSO+trehalose. After 12 months of cryopreservation cells were assessed for viability, differentiation, proliferation and pro-inflammatory and anti-inflammatory cytokine markers.

RESULTS: CS+DMSO had the ($p < 0.05$) greatest average viability (83%), excluding FBS+DMSO (78%). Trehalose+DMSO viability (65%) was statistically different from all except FBS+DMSO. All other conditions had $< 40\%$ viability. DMSO allowed consistent proliferation and differentiate; those lacking DMSO could not. Expression of pro-inflammatory markers was significantly less ($p < 0.05$) in FBS alone compared to DMSO+FBS. CS and FBS had significantly lower expression of COX2 markers than DMSO+FBS or DMSO+CS. Trehalose+DMSO had significantly higher COX2 marker levels than DMSO+FBS or DMSO+CS. Conditions with DMSO had significantly ($p < 0.05$) higher concentrations of TIMP-2 than those without DMSO.

DISCUSSION/CONCLUSION: Viability, differentiation and proliferation is dependent upon DMSO. DMSO is associated with higher levels of pro-inflammatory and anti-inflammatory cytokine levels, therefore we reject our hypothesis. CS and trehalose show promise as adjunctive cryoprotectant substitutes for FBS.

ACKNOWLEDGEMENT: qRT-PCR performed by the University of Minnesota Genomics Center.

38 Allogeneic Stem Cell Survival and Localization after Intra-articular Administration in Laboratory Beagles

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INTRODUCTION: Methods of evaluating cell biodistribution utilize radio-labeling or tagging with materials such as iron nanoparticles. Such methods can alter cell migration and viability. Use of sex-mismatched cells with a PCR detection assay can avoid such artifacts. This study determined the survival and tissue biodistribution of allogeneic adipose-derived stem cells (ASC) after intraarticular administration in laboratory beagles.

MATERIALS & METHODS: The ASCs were derived from a male donor dog, providing Y-chromosome DNA to be injected into female recipients. The DNA was detected via a PCR assay at a sensitivity of 1 cell in 10,000. Twelve dogs were injected with ASCs on Day 0. Two dogs were negative controls. On each collection day (15, 30, 60, 90, 120) there were two ASC dogs sacrificed and two control dogs were sacrificed on day 15. Eighteen tissue samples were taken at each sacrifice.

RESULTS: The control dogs showed no detectable male cells. In ASC-treated dogs, no cells were detected outside of the injected joints. ASCs were detected in the cartilage of two dogs at Day 15 and one dog at Day 60.

DISCUSSION/CONCLUSION: The ASC dogs showed cell engraftment in cartilage out to day 60 and no detectable systemic distribution. This confirms the actual cell survival and distribution within the limits of this assay.

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39 Inter- and Intra-Observer Variability of Radiography and Computed Tomography for Evaluation of Zurich Cementless Acetabular Cup Placement Ex Vivo

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INTRODUCTION: Total hip arthroplasty (THA) is a well-established technique for surgical management of end-stage osteoarthritis of the coxofemoral joint. Accurate intra-operative positioning of the acetabular component of THA is exceptionally challenging. The purpose of this ex vivo cadaveric study was to evaluate inter- and intra-observer variability in measurement of angle of lateral opening (ALO) and version angle for acetabular cup position using digital radiography and computed tomography.

MATERIALS & METHODS: Each hemipelvis was implanted with a cementless acetabular cup. Ventrodorsal and mediolateral radiographs were made of each pelvis, followed by CT. The pelvis were explanted and re-implanted with an acetabular cup in the contralateral acetabulum for repeated imaging. Three surgeons measured the ALO and version angles three times for each cup using the mediolateral radiographic projection. The same measurements were made using three-dimensional multiplanar CT images. Two-way repeated measures analysis of variance evaluated inter- and intra-observer repeatability for both imaging modalities.

RESULTS: Radiographic version angle measurement did not differ within surgeons ($p = 0.433$), but differed between surgeons ($p < 0.001$). Radiographic measurement of ALO differed within surgeons ($p = 0.006$) but not between surgeons ($p = 0.989$). The ALO and version angle measured on CT images did not differ with or between surgeons.

DISCUSSION/CONCLUSION: Assessment of inter- and intra-observer measurement of ALO and version angle was more reproducible using CT than conventional radiography for a Zurich cementless acetabular cup.

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40 Kinetic Gait Analysis of Agility Dogs Entering the A-Frame

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INTRODUCTION: To investigate the effect of a decrease in the inclination angle of the A-frame on the peak vertical force (PVF), heel strike transient (HST) and the rate of force development to the PVF and HST (R-PVF, R-HST) in agility dogs.

MATERIALS & METHODS: A randomized, factorial study design of A-frame angles 40°, 35° and 30° was applied to 26 agility dogs. Kinetic gait analysis measuring the PVF, HST and R-PVF, R-HST was performed in dogs entering the A-frame at all three angles of inclination. Kinetic output was normalized to body weight and gravity. Statistical analysis included the covariates height and velocity.

RESULTS: There was no significant difference in the normalized kinetic output between the A-frame angles. The mean velocity was 5.3 m/s. The normalized least square mean at the standard angle of inclination (40°) for the PVF was 2.3N/kg and for the HST it was 1.3N/kg.

DISCUSSION/CONCLUSION: A decrease in the angle of the A-frame did not result in lower PVF, HST, R-PVF, or R-HST. Whether this magnitude of PVF relates to injury risk is unknown but reducing the A-frame angle is unlikely to make a difference.

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41 Variability of the Position of the Greater Trochanter: Implications for Femoral Preparation during Canine Total Hip Replacement

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INTRODUCTION: Femoral broaching in cementless total hip replacement (THR) should be coaxial to the femoral long axis. The medial border of the greater trochanter (GT) is considered a lateral landmark for femoral preparation, but its position with regard to an ideal femoral preparation is variable.

In this study, radiographic measurement of the position of the GT was validated by comparison to CT. Position of the GT was then measured on radiographs in a population of dogs.

MATERIALS & METHODS: Radiographic measurements were compared to an anatomic standard (CT) in 56 femurs. The lateral margin of the femoral envelop was defined by a line parallel to the femoral long axis, and tangential to the lateral endosteal surface at the femoral isthmus. The mediolateral displacement of the medial border of the GT from this line was measured on radiographs of 233 femurs.

RESULTS: There was no difference between CT and radiographic measurements ($p = 0.54$). 41% (96/233) of femurs had medialized GT (range 6.9 mm medial to 10.9 mm lateral). Dogs with hip dysplasia were significantly more likely to have GT medialization ($p = 0.01$; OR = 4.12; 95%CI = 1.4–11.8) as were German Shepherd Dogs ($p = 0.00$; OR = 3.86; 95%CI = 1.87–7.95). Golden Retrievers were less likely to have medialization ($p = 0.05$; OR = 0.44; 95%CI = 0.2–0.98).

DISCUSSION/CONCLUSION: Radiographic determination of displacement of the GT is accurate. There is wide variability in position of the GT. If the GT is medialized, depletion of the GT may be necessary during broaching for proper stem alignment. Routine alignment of the terminal femoral broach with the medial border of the GT is not advisable.

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42 Effect of Regional Limb Perfusion Volume on Concentrations of Amikacin Sulfate in Synovial and Interstitial Fluid Samples from Anesthetized Horses

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INTRODUCTION: To evaluate the effect of volume of intravenous regional limb perfusion (IVRLP) on amikacin concentrations of metacarpophalangeal joint and metacarpal interstitial fluid in anesthetized horses.

MATERIALS & METHODS: Eight horses were used, each fore limb was randomly assigned to receive a large volume (60 mL) or small volume (10 mL) IVRLP. Under general anesthesia, IVRLP was performed using the lateral palmar digital vein. The tourniquet remained in place for 30 minutes. Venous blood pressure of the distal limb was recorded throughout the procedure. Synovial and interstitial fluid samples were obtained at times 0, 30 min (before tourniquet removal), and 24 hours and 0, 6 hours, and 24 hours, respectively.

RESULTS: Synovial amikacin concentration at 30 min for the large volume (mean 459 µg/mL) was significantly greater than for the small volume (mean 70 µg/mL). Interstitial amikacin concentrations at 6 hours for the large volume (mean 723 µg/mL) was significantly greater than for the small volume (mean 21 µg/mL). Peak venous pressures following large volume IVRLP were significantly higher than for small volume IVRLP, but there was no difference between treatments in time required for venous pressures to return to baseline.



DISCUSSION/CONCLUSION: The large volume IVRLP resulted in significantly greater synovial and interstitial amikacin concentrations than small volume IVRLP. Although venous pressures were higher in the large volume, those pressures were not sustained.

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43 Incidence of Radiographic Patellar Tendon Thickening Following Tibial Plateau Leveling Osteotomy Using Three Methods of Joint Inspection

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INTRODUCTION: Radiographic patellar tendon thickening (PTT) and clinical patellar desmitis are reported complications following tibial plateau leveling osteotomy (TPLO). The objective of this study was to determine the incidence of radiographic PTT after TPLO when joints were examined with arthroscopy (AR), via craniomedial arthrotomy (CrMA), or caudomedial arthrotomy (CdMA). Our hypothesis was that method of joint inspection influences PTT and that the incidence of PTT would be lower following arthroscopy.

MATERIALS & METHODS: Medical records of 210 dogs with CCL rupture undergoing 218 TPLOs were reviewed for data on signalment, weight, anesthesia time, and surgery time. Radiographs were reviewed by a blinded radiologist, and patellar tendon thickness was measured pre-operatively, post-operatively, and at 10 weeks post-operatively. Cases which converted from arthroscopy to CrMA were considered a separate group (CA). Grades (0, 1, 2) were assigned, with grades 1 and 2 considered to have mild and severe PTT, respectively.

RESULTS: The AR and CA groups had significantly longer surgery and anesthesia times, greater post-operative PTT, recheck PTT, and combined grade 1 and 2 PTT (78.04%, 89.47%) compared to CdMA (28.17%) and CrMA (52.87%). The incidence of combined grade 1 and 2 PTT was significantly greater after CrMA compared to CdMA.

DISCUSSION/CONCLUSION: The incidence of radiographic PTT was significantly greater for joints inspected with AR or CA compared to CrMA or CdMA. Joint approach influences the development of PTT following a TPLO, with potential to contribute clinical patellar desmitis.

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44 Changes in the Flexor Tendons Following Injury: An Ex-Vivo Study Using an Ovine Tendinopathy Model

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INTRODUCTION: Pathological changes are observed throughout the entire tendon following focal flexor tendinopathy; however, little is known about potential changes in adjacent tendons following injury. The aim of this study was to investigate the gene expression and histopathological changes that occur within the deep digital flexor tendon (DDFT) after injury to the superficial digital flexor tendon (SDFT) and the flexor tendons (SDFT and DDFT) after injury to the extensor tendons.

MATERIALS & METHODS: Merino wethers (n=18) were split into three groups and underwent either partial transection of the SDFT, complete transection of the extensor tendons or left as non-operated controls. Tendons were harvested and regionally sampled for gene expression (real time PCR) and histologic analysis at 8 weeks postoperatively.

RESULTS: Transection of the SDFT resulted in increased expression of collagen III, versican, biglycan and lumican (P<0.026 for all genes) within the DDFT. In particular, lumican expression was markedly increased (up to 220 fold). There was no effect of transecting the extensor tendons on the expression of any gene tested in either the SDFT or the DDFT.

Transection of the SDFT increased the histopathology scores in the DDFT. There were no histological differences in either the SDFT or DDFT after transection of the extensor tendons.

DISCUSSION/CONCLUSION: Transection of the SDFT results in a focal, subclinical tendinopathy within the DDFT with potential implications on treatment and rehabilitation of SDFT injuries. Injury to the extensor tendons has no effect on the SDFT or DDFT.

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45 Comparison of Outcome Following Semitendinosus Tenotomy Performed Under Standing Sedation Versus General Anesthesia for the Treatment of Fibrotic Myopathy in Horses

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INTRODUCTION: This retrospective study reports the outcome following a modified technique for semitendinosus tenotomy, performed under standing sedation, as compared to those performed under general anesthesia (GA); and reports outcome following semitendinosus tenotomy overall.

MATERIALS & METHODS: Medical records (2002–2015) of horses undergoing semitendinosus tenotomy for the treatment of fibrotic myopathy under standing sedation and GA were retrospectively reviewed. Long-term outcome was subdivided into 4 groups based on gait improvement and post-operative athletic use. Outcome was established via follow-up contact with owners.

RESULTS: Overall, 62% of cases benefited to varying degrees from semitendinosus tenotomy surgical intervention. Of 8 horses treated under standing sedation, 37.5% had full improvement of gait, 25% had moderate improvement with mild residual gait deficit, 12.5% had mild improvement with moderate residual gait deficit, and 25% had no improvement. Of the 13 limbs of 12 horses treated under GA, 23% had full improvement in the gait, 23% had moderate improvement with mild residual gait deficit, 8% had mild improvement with moderate residual gait deficit and 46% had no improvement. Overall, 83% of horses treated under standing sedation and 67% treated under GA were able to return to the same level of pre-injury athletic performance.

DISCUSSION/CONCLUSION: Performing semitendinosus tenotomy under standing sedation, as compared to GA, allows for easier palpation of the semitendinosus tendon insertion, has reduced cost, and results in similar outcomes for improving the gait deficit and return to athletic performance.

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